

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions and listings of claims in the application:

LISTING OF CLAIMS:

1-12. (canceled).

13. (currently amended): A method for producing a biomolecule, whereby about 95% or more of the atoms in the biomolecule, for at least one of H, C or N, are isotopically labelled, the method comprising the steps of:

(a) growing an organism on a mineral medium which supports growth of the organism, whereby in the medium about 95% or more of the assimilable atoms, for at least one of H, C or N are isotopically labelled, to produce labelled biomass;

(b) autolysing the biomass of the organism grown as in (a) to produce an autolysate;

(c) composing the nutrient medium by combining the autolysate as obtained in (b) with further components necessary for growth of the mammalian or insect cells;

(d) growing a culture of mammalian or insect cells producing the biomolecule under conditions conducive to the production of the biomolecule, in a~~the~~the nutrient medium produced by:

(i) growing an organism on a mineral medium which supports growth of the organism, whereby in the medium about 95% or more of the assimilable atoms, for at least one of H, C or N, are isotopically labelled, to produce labelled biomass;

(ii) autolysing the biomass of the organism grown as in (i) to produce an autolysate; and,

(iii) composing the nutrient medium by combining the autolysate as obtained in (ii) with further components necessary for growth of the mammalian or insect cells; and

~~(b)(e)~~ recovery of the biomolecule,

whereby the biomolecule is a molecule that is naturally synthesized by the mammalian or insect cells, or the biomolecule is a mammalian polypeptide or nucleic acid produced as a result of genetic engineering of the mammalian or insect cells.

14. (original): A method according to claim 13, wherein the biomolecule is a membrane protein.

15. (original): A method according to claim 14, wherein the mammalian or insect cells comprise an expression vector comprising a nucleotide sequence coding for the protein from which the protein is produced.

16. (canceled).

17. (withdrawn): A method for obtaining structural information on a biomolecule, the method comprising the steps of:

(a) producing a biomolecule, whereby substantially all atoms in the biomolecule are isotopically labelled, in a method according to ;
(b) optionally, purifying the biomolecule;
(c) subjecting the biomolecule to spectroscopic analysis to obtain information about its structure.

18. (withdrawn): A method according to claim 17, wherein the spectroscopic analysis comprises NMR spectroscopy.

19. (withdrawn): A method according to claim 17, wherein the structural information on a biomolecule is information about the three-dimensional structure of the biomolecule.

20. (withdrawn): A method according to claim 17, wherein the biomolecule is a protein complexed to a second biomolecule.

21. (withdrawn): A method according to claim 20, whereby 20 - 100% of the hydrogen atoms in the second biomolecule are uniformly substituted with the isotope ^2H .

22. (withdrawn): A method according to claim 21, wherein the second biomolecule is a protein.

23. (withdrawn): A nutrient medium for the production of an isotopically labelled biomolecule from mammalian or insect cells, the medium supporting growth of a mammalian or insect cell culture under condition conducive to the production of the biomolecule, the medium comprising:

(a) a mixture of inorganic salts;

(b) a source of amino acids;

(c) a carbohydrate energy source;

(d) a source of lipids;

(e) optionally, a protective agent;

(f) optionally, vitamins and/or organic compounds;

(g) optionally, organic acids; and,

(h) optionally, trace elements;

whereby substantially all atoms in (a), (b), and (c), and, optionally in (d), (e), (f), (g) and (h) are isotopically labelled for at least one of H, C or N or whereby 20 - 100% of the hydrogen atoms in (a), (b), and (c), and, optionally in (d), (e), (f), (g) and (h) are uniformly substituted with the isotope ^2H .

24. (withdrawn): A nutrient medium according to claim 23, whereby the source of amino acids comprises an hydrolysate comprising amino acids that is produced from yeast biomass, whereby the hydrolysis of the biomass comprises autohydrolysis.

25. (withdrawn): A nutrient medium according to claim 23, whereby the source of lipids comprises fatty acids, steroids, and lipid soluble vitamins.

26. (withdrawn): A nutrient medium according to claim 23, whereby the carbohydrate energy source is one or more of glucose, fructose, and sucrose; the organic acids are one or more of pyruvate and the Krebs-cycles intermediates selected from the group consisting of citrate, succinate, fumarate, maleic acid, oxalate and malate; the vitamins are one or more vitamins selected from the group consisting of thiamin, riboflavin, niacin, vitamin B6, folic acid, vitamin B12, biotin, pantothenic acid, choline, para-aminobenzoic acid and alpha-tocopherol.

27. (withdrawn): A nutrient medium according to claim 23, whereby substantially all atoms in (a), (b), and (c), and, optionally in (d), (e), (f), (g) and (h) are isotopically labelled with an isotope selected from ^{15}N ; ^{13}C ; ^2H ; ^{15}N and ^{13}C ; ^{15}N and ^2H ; ^{13}C and ^2H ; or ^{15}N , ^{13}C and ^2H .

28. (withdrawn): A mammalian membrane protein whereby substantially all atoms in the protein are isotopically labelled with an isotope selected from ^{15}N , ^{13}C ^2H ^{15}N and ^{13}C , $'\text{N}$ and ^2H , ^{13}C , and ^2H , or ^{13}C and ^2H .

29. (withdrawn): A mammalian membrane protein whereby 20 - 100% of the hydrogen atoms in the protein are uniformly substituted with the isotope ^2H .

30. (withdrawn): A mammalian membrane protein according to claim 28, whereby the protein is a human protein.

31. (previously presented) A method according to claim 13, wherein the organism is a fungus, yeast or algae.

32. (previously presented) A method according to claim 31, wherein the organism is an organism that belongs to a genus selected from *Saccharomyces*, *Pichia*, *Hansenula*,

Kluyveromyces, Candida, Brettanomyces, Debaryomyces, Tolrulopsis, Yarrowia, Galdieria, Cyanidium, Porphyridium, Cystoclonium, Audouinella, and Cyanidioschyzon.

33. (currently amended) A method according to claim 13, wherein the method for producing the nutrient medium further comprises the steps of:

(iv)(i) growing ~~an-a second~~ organism on a mineral medium which supports growth of the ~~second~~ organism, whereby in the medium about 95% or more of the assimilable atoms, for at least one of H, C or N, are isotopically labelled, to produce labelled biomass;

(v)(ii) extracting biomass of ~~an-the second~~ organism ~~grown in (i)~~ with an organic solvent to produce an extract comprising lipids, ~~whereby the organism is grown as in (iv) or is grown as in (iv) on a medium without isotopic substitution;~~

(vi)(iii) hydrolysing biomass of ~~an-the second~~ organism grown as in (iv)(ii) at a non-alkaline pH to produce a hydrolysate comprising amino acids; and,

(vii)(iv) composing the nutrient medium by combining the autolysate obtained in (b) with amino acids as obtained in (vi)(iii) and the lipids obtained in (v)(ii) and adding further components necessary for growth of the mammalian or insect cells.

34. (previously presented) A method according to any one of claim 33, whereby the nutrient medium is composed of autolysate, lipids and amino acids obtained from at least two different organisms.

35. (previously presented) A method according to claim 33, whereby, prior to hydrolysis in (vi), lipids and pigments are extracted from the biomass using an organic solvent.

36. (previously presented) A method according to claims 33, whereby the organism from which the lipids are extracted, belongs to a genus selected from the group consisting of *Rhodophyta, Cyanidiophyceae, Chlorophyta, Cyanophyta, Diatoms, Phaeophyceae, Dinoflagelate, Dinophyta and Galdieria.*

37. (currently amended) A method according to claim 33, whereby the organism from which the hydrolysate comprising amino acids is produced, is an organism selected from the group consisting of algae, fungi, yeasts and methylotrophic bacteria.

38. (previously presented) A method according to claim 37, whereby the organism belongs to a genus selected from the group consisting of *Pichia*, *Saccharomyces*, *Hansenula*, *Cyanidium*, *Galdieria*, *Porphyridium*, *Spirulina*, and *Methylobacillus*.

39. (previously presented) A method according to claim 13, whereby the further components necessary for growth of the mammalian or insect cells comprise one or more of:

- (a) one or more of glucose, fructose, and sucrose;
- (b) one or more Krebs-cycles intermediates selected from the group consisting of citrate, succinate, fumarate, maleic acid, oxalate and malate;
- (c) pyruvate; and,
- (d) one or more vitamins selected from the group consisting of thiamin, riboflavin, niacin, vitamin B6, folic acid, vitamin B12, biotin, pantothenic acid, choline, para-aminobenzoic acid and alpha-tocopherol.

40. (previously presented) A method according to claim 13, whereby about 95% or more of the atoms in substrates that are used by the mammalian or insect cells for synthesis of biomolecules in the nutrient medium are isotopically labelled with an isotope selected from ^{15}N ; ^{13}C ; ^2H ; ^{15}N and ^{13}C ; ^{15}N and ^2H ; ^{13}C and ^2H ; or ^{15}N , ^{13}C and ^2H .

41. (new) A method according to claim 13, wherein the method for producing the nutrient medium further comprises the steps of:

- (i) growing a second organism on a mineral medium which supports growth of the second organism, whereby in the medium about 95% or more of the assimilable atoms, for at least one of H, C or N, are isotopically labelled, to produce labelled biomass;

- (ii) extracting biomass of a third organism with an organic solvent to produce an extract comprising lipids, whereby the third organism is grown on a medium without isotopic substitution;
- (iii) hydrolysing biomass of the second organism grown in (i) at a non-alkaline pH to produce a hydrolysate comprising amino acids; and,
- (iv) composing the nutrient medium by combining the autolysate obtained in (b) with amino acids as obtained in (iii) and the lipids obtained in (ii) and adding further components necessary for growth of the mammalian or insect cells.